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THE ROLE OF FLAGELLATES IN PIONEER PROTOZOAN COLONIZATION OF ARTIFICIAL SUBSTRATES

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ABSTRACT

In earlier studies using polyurethane foam for protozoan colonization, communities were established in 14 days (sampling weekly). Daily sampling in this study revealed that the number of colonizers increases with time and that flagellates make up the major portion of the early pioneer community becoming established in two to three days and reaching equilibrium much earlier than other taxonomic groups. Additional data indicate the flagellate component having reached equilibrium remains relatively stable during seasonal changes for as long as two years. A list of most common summer pioneer and persistant flagellates in two kinds of ecosystems is given.

1. INTRODUCTION

In an earlier study (Yongue, Cairns 1971) results indicated that of 96 freshwater protozoan species which colonized polyurethane foam substrates over a period of 18 weeks, 30.2% were present by the end of two weeks. These pioneer colonizers also represented an average of 54.8% of the species ("resident species") that persisted throughout the study period. This period was from late December to early June and was characterized by significant changes in water temperature and other water quality characteristics. It is significant that the total number of species present in a previously unsampled polyurethane unit was never higher than 37 species, and this particular instance occurred after 14 weeks. The number of species at the end of 14 days (2 weeks) was 21, and the average for the total period was 25.77 species (SD=7.26). This suggests that a dynamic equilibrium was reached by the end of 14 days.

During the study just described, sampling was carried out every seventh day. Having no knowledge of pioneer colonization, with the exception of the two points—day 7 and day 14—the pattern of early colonization during this period was not evident. There were the usual oscillations about the mean after the second week. The opportunity to make observations on early colonization patterns was presented during the summers of 1973 and 1974 as a part of the RANN Project (National Science Foundation RANN Grant No. GI-34898) being carried out by the University of Michigan Biological Station.

The protozoan portion of this investigation was specifically designed to observe the pattern of colonization on polyurethane foam by freshwater protozoans during the pioneer phase.

2. MATERIALS AND PROCEDURES

During the early summer of both 1973 and 1974, sets of polyurethane foam units (PF units or units) were anchored under approximately 0.5 meter of water in the epilimnion of South Fishtail Bay of Douglas Lake, Cheboygan County, Michigan. The units measured 76 mm imes 64 mm imes 51 mm. They were tied by nylon cord to weighted plastic bags so that they could not directly contact the natural substrate and were then distributed in a random fashion about the selected area. Subsets of 3 units were subsequently harvested after having been exposed to the lake for 1 to 10 days. Harvesting involved cutting a unit from its plastic bag anchor and inserting it into a wide mouth jar partly filled with lakewater from the area where it was collected. Although the lakewater was not protozoan free, it contained low density planktonic species quite different from those associated with the substrate. The harvested units were promptly taken to the laboratory at the University of Michigan Biological Station and sampling was begun within one hour after collection. Each unit was taken from the harvesting jar and the material in it was squeezed into a small wide mouth jar. Four wetmount slides, each consisting of four drops of material taken from the sample jar, were examined under the microscope. The kinds of protozoan species present were recorded. This procedure was much the same as that reported in earlier papers (Cairns et al. 1971, Yongue, Cairns 1971). Standard protozoology keys were used for making identifications. Limited water quality determinations were made at the sampling site using a Hach Kit and a thermometer.

The results obtained were compared with results from several other Douglas Lake studies. Specifically, the total species colonizing the units and the numbers of species of the classes were reduced to percentages and plotted. These comparisons included colonization of units that had been exposed in the lake in the same general area for periods of two years, one year, three months, three weeks, and nine days over seasonal periods including early summer, late summer, and early fall after lake overturn. In addition, colonization information from a soft-water, low pH, eutrophic bog pond and from North Fishtail Bay of Douglas Lake was used for contrast with the sampling in the South Fishtail Bay area where most of our studies have been carried out.

3. RESULTS

Limited water quality determinations were made during the course of this study and the results are given in Tab. I. The periodic examinations made from 4 July through 27 July 1973 in South Fishtail Bay of Douglas Lake showed minor fluctuations in quality. The temperature changed by only 2°C (between 22 and 24°C), pH remained at 8.8 (except one occasion when it was 9.0), total hardness remained between 153.9 and 136.8, and dissolved oxygen concentration was as low as 7 ppm and as high as 10 ppm. There was no substantial difference between these determinations and those that were made a year later during the period 26 June—15 July 1974. The temperature rose from 21 to 24°C, pH remained at 8.8 throughout the period as total hardness remained at 153.9 to 136.8 ppm, and dissolved oxygen concentration was 9 ppm except at the beginning and the end when it was 8 ppm. In 1974 there was an Anabaena bloom from 26 June to 29 June overlapping a Dinobryon bloom (29 June—8 July) that overlapped a Tabellaria bloom (8 July—15 July).

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Table I. Water quality determinations

			I. D	ouglas Lake	e, South Fis	shtail Bay —	1973		
Date	4 July	9 July	12 July	14 July	16 July	21 July	27 July		
Temperature (°C)	24	25	22	22	22	23	23		
pH	8.8	8.8	8.8	8,8	8.8	9.0	8.8		
Hardness (total, ppm)	153.9	136.8	136.8	136.8	153.9	153.9	153.9		
Dissolved oxygen (ppm)	10	9	8	7	8	8	8		
			II. D	ouglas Lake	, South Fis	htail Bay —	1974		
Date	26 June	29 June	30 June	3 July	5 July	6 July	8 July	12 July	15 July
Temperature (°C)		21	21	21	20	21		23	24
pH		8.8	8.8	8.8	8.8	8.8	-	8.8	8.8
Hardness (total, ppm)		153.9	153.9	153.9	153.9	153.9		153.9	153.9
Dissolved oxygen (ppm)		8	9	9	9	9		9	8
Anabaena bloom	*	*							
Dinobryon bloom	- 0	*	*	*	*	*	z)e		
Tabellaria bloom							*	*	sje
				III. Niche	ol's Bog Pon	d — 1970		2	
Date	23 June	30 June	7 July	14 July	21 July	28 July	4 Aug	11 Aug	
Temperature (°C)	21	24	23	24	18	26	. 20	24	
pH	5.8	5.8	6.0	5.8	5.8	6.0	6.0	6.0	
Hardness (total, ppm)	17.1	17.1	34.2	17.1	< 17.1	17.1	34.2	17.1	
Dissolved oxygen (ppm)	10	6	8	7	7	7	6	6	
				IV. Nicho	ol's Bog Pone	d — 1974			
Date	2 July	9 July	16 July	30 July	6 Aug	13 Aug			
Temperature (°C)	21	24	21	20	21	21			
pH	5.8	5.5	6.0	5.5	6.0	6.0			
Hardness (total, ppm)	< 17.1	< 17.1	< 17.1	< 17.1	< 17.1	< 17.1			
Dissolved oxygen (ppm)	7	8	7	6	8	8			

No sample taken.

Observed.

Table I also contains water quality determinations from Nichol's Bog Pond, Cheboygan County, Michigan, which has an open-water edge that borders a dairy cattle lot and is used by the cattle for drinking. From 23 June—11 August 1970 the water temperature fluctuated from 21°C at the beginning to 18°C on 21 July with the highest of 26°C on 28 July. With two exceptions the total hardness was 17.1 ppm or less, the pH ranged between 5.8 and 6.0, and the average dissolved oxygen concentration was about 7 ppm with a range from 10 ppm to 6 ppm. In 1974, during the 2 July—13 August period, temperature ranged between 20 and 24°C, pH between 5.5 and 6.0, total hardness was always less than 17.1 ppm, and dissolved oxygen concentration ranged from 8 ppm to 6 ppm.

Table II. Pioneer colonization of polyurethane foam units by freshwater protozoans (Douglas Lake, South Fishtail Bay; started 27 June 1973)

Days	Unit 1	Unit 2	Unit 3	Means
Exposed	FSCT	F S C T	F S C T	F S C T
1	4 1 2 7	8 1 6 15	5 1 1 7	5.7 1.0 3.0 9.7
2	21 5 16 42	18 15 17 50	15 8 16 39	18.0 9.3 16.3 43.7
3	12 5 14 31	15 5 12 32	11 2 9 22	12.6 4.0 11.7 28.3
4	15 5 11 31	17 10 19 46	12 8 13 33	14.7 7.6 14.3 36.7
5	12 4 6 22	9 6 2 17	15 6 9 30	12.0 5.3 5.7 23.0
6	17 6 12 35	20 8 11 39	12 11 12 35	16.3 8.3 11.7 36.3
7	14 9 16 39	18 12 15 45	17 10 18 45	16.3 10.3 16.3 43.0
8	16 10 17 44	18 7 16 41	15 9 12 36	16.3 8.6 15.0 40.3
9	21 11 19 51	18 17 22 57	15 11 27 59	18.0 13.0 22.7 55.7
10	18 14 26 57	17 10 25 52		17.5 12.0 25.5 54.5
14	14 8 29 51	15 11 17 43	15 17 29 61	14.7 12.0 25.0 52.0
14	14 12 22 47	6 13 22 41	10 16 26,52	10.0 13.6 23.3 46.7
14	11 15 26 52	9 20 26 54	14 13 14 41	11.3 16.0 22.0 49.0

F - Flagellates,

Table II shows the numbers of freshwater protozoans colonizing polyurethane foam units in 1973 during the pioneer period from 1 day of exposure to 10 days and 14 days in South Fishtail Bay. Based on sets of 3 units sampled, the mean number of protozoan species rose from 9.7 after 1 day exposure to 43.7 on day 2. The highest mean number of species was obtained on day 9 (55.7) with those (3 sets) on day 14 being slightly lower (52, 46.7 49). It should be noted that the mean for day 10 is based on 2 units instead of 3 since 1 unit was inadvertently lost.

Table III gives the results for the same sampling protocol as in Tab. II approximately one year later. The mean number of species (20.3) on the first day was more than twice as great as the year before, and on the second day, the mean was about the same as day 1 which was about half as great as day 2 on the year before. This suggests that invasion rates are not uniform or that the substrates are so small that wide

S - Sarcodines,

C - Ciliates.

T - Total species observed,

⁻ No sample taken.

Table III. Pioneer	colonization of polyurethane foam units by freshwater protozoans (Douglas
14010 1111 110110	Lake, South Fishtiail Bay; started 5 July 1974)

Dove	Unit 1	Unit 2	Unit 3		Mea	ans	
Days Exposed	FSCT	FSCT	FSCT	F	S	С	T
1	16 1 8 25	15 2 3 20	11 1 4 16	14	1.3	5	20.3
2	14 3 9 26	12 0 6 18	11 5 5 21	12.3	2.7	6.7	21.7
3	15 4 10 29	14 4 12 30	15 7 10 32	14.7	5	10.7	30.3
4	12 9 14 35	13 7 19 39	25 9 12 46	16.7	8.3	15	40
5	10 6 12 28	8 10 6 24	10 6 10 26	9.3	7.3	9.3	26
6	17 7 15 39	14 11 16 41	14 9 13 36	15	9	14.7	38.7
7	11 10 19 40			11	10	19	40
8	16 10 16 42	15 15 18 48	14 12 19 45 .	15	12.3	17.7	45
9	14 11 16 41	15 4 11 30	13 9 12 34	14	8	13	35
10	19 14 17 50	17 11 16 44	13 12 19 44	16.3	12.3	17.3	46
14	12 11 18 42	14 12 18 46	15 14 23 52	13.6	12.3	19.6	46.7

F - Flagellates,

variations in the number of viable species are likely to occur in the early stages of colonization. In general, the total numbers of species were slightly lower in 1974 than the year earlier but were well within the 1973 ranges. Only one unit was sampled on the seventh day. The total number of species per unit or per set show an increasing trend during both years. Calculation of rank correlation gives S=23, r=0.51 for 1973 and S=30, r=0.66 for 1974 significant at $0.23^{\circ}/_{\circ}$ and $0.35^{\circ}/_{\circ}$, respectively. In addition, species richness on day 14 appears to be comparable in both years (Figs. 1 and 2).

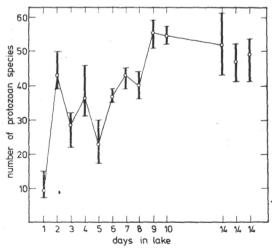


Fig. 1. Pioneer colonization of polyurethane foam by freshwater protozoans — early summer 1973, Douglas Lake, Michigan. Started: 27 June 1973. Points — mean values, vertical lines — range

S - Sarcodines,

C - Ciliates,

T -- Total species observed,

⁻ No sample taken.

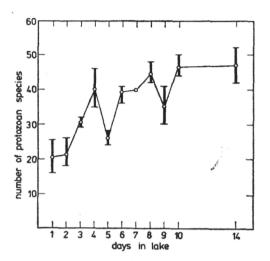


Fig. 2. Pioneer colonization of polyurethane foam by freshwater protozoan — early summer 1974, Douglas Lake, Michigan. Started: 5 July 1974. Mean values and range are shown

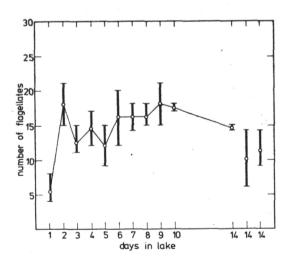


Fig. 3. Flagellates among pioneer protozoan colonizers — early summer 1973, Douglas Lake. Started: 27 June 1973. Mean values and range are shown

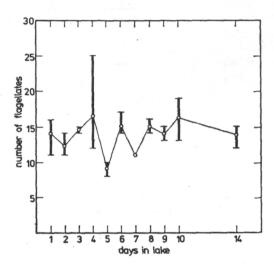


Fig. 4. Flagellates among pioneer protozoan colonizers — early summer 1974, Douglas Lake. Started: 4 July 1974. Mean values and range are shown

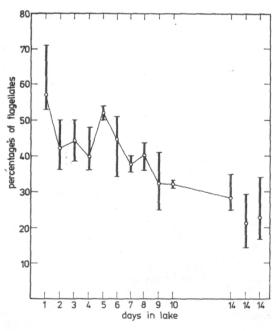


Fig. 5, Percentages of flagellates in PF units — early summer 1973, Started: 27 June 1973, Mean values and range are shown

Figures 3 and 4 show that the flagellates identified from the PF units in 1973 acquired a stable species number quite early in the colonization process and that this number did not increase appreciably,

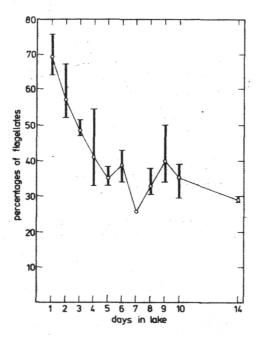


Fig. 6. Percentages of flagellates in PF units — early summer, 1974. Started: 5 July 1974. Mean values and range are shown

except for short-term fluctuations, after that. Figures 5 and 6 show the percentage range and means of number of flagellate species in the total species pool for sets of three units per sampling day for 1973 and 1974, respectively. In both cases, by day 14 the percentage of flagellates has dropped from more than $50^{\circ}/_{\circ}$ to $35^{\circ}/_{\circ}$ or less.

Table IV shows the percentage of flagellates colonizing units from multiple sets that were placed in South Fishtail Bay of Douglas Lake for various exposure times and during different seasons. The first set, sampled 28 June 1974, contained 10 units that had remained in the lake for 2 years. The percentage of protozoans colonizing the PF units that were flagellates ranged from $13.6^{\circ}/_{0}$ to $33.3^{\circ}/_{0}$ with a mean of $20.54^{\circ}/_{0}$. In the set that had been exposed for 1 year, the range was $11.1^{\circ}/_{0}$ to $30^{\circ}/_{0}$ with a mean of $19.07^{\circ}/_{0}$. A third set with an exposure to the lake of three months was sampled 5 October 1974. This date was one week after lake overturn. The percentage of flagellates in PF units ranged from $21.2^{\circ}/_{0}$ to $29.7^{\circ}/_{0}$ with a mean of $25.6^{\circ}/_{0}$. The mean total number of protozoan species was 60.6 and for flagellates it was 15.5.

At the time that the 3-month-old set was harvested and sampled, a previously unexposed set was placed in the lake. Three weeks later, 26 October 1974—four weeks after lake overturn, this set was harvested

Table IV. Percentages of flagellates colonizing polyurethane foam units in multiple sets (South Fishtail Bay, Douglas Lake, Michigan)

Exposure	Sampling date	Unit number	Total flagellates	Total protozoan species	% flagellates
2 years	28 June 1974	1	6	34	17.6
1.		2	3	17	17.6
t		3	7	33	21.2
		4	6	18	33.3
•		5	6	33	18.2
* i		6	9	46	19.6
		7	6	28	21.4
: *.		8	3	22	13.6
		9	7	33	21.2
		10	5	23	21.7
l year		1	15	50	30
		. 2	13	46	28.3
		3	12	45	26.7
•		4	9	41	21.9
		5	6	45	13.3
•		. 6	4	36	11.1
	-	7	5	35	14.3
		8	6 -	40	15
.***		9	4	36	11.1
		10	8	42	19
3 months	5 October 1974	1	19	64	29.7
(1 week after		2	13	47	27.7
lake overturn)		3	19	71	26.8
		4	18	68	26.5
		5	13	58	22.4
		6	18	72	25
		7	14	66	21.2
7 7 7		8	12	51	23.5
		9	14	54	25.9
		10	15	55	27.3
3 weeks	26 October 1974	1	17	47	36.2
(4 weeks after		2	7	35	20
lake overturn)		3	10	41	24.4
		4	8	34	23.5
		5	7	41	17.1
		6	16	59	27.1
		7	16	56	28.6
	. %.	8	10	46	21.7
			12	48	27.1
		9	13		
		10	9	41	21.9

Exposure	Unit number	Sampling date	Total flagellates	Total protozoan species	% flagellates
9 days	11 July 1970	1	19	35	54.3
		2	15	31	48.4
		3	15	32	46.8
		4	14	28	50
		5	13	33	39.4
	-	6	14	36	38.9
		7	13	31	41.9
		8	17	38	44.7
		9	10	30	33.3
		10	14	36	38.8

and examined. The percentage of flagellates among the colonizing protozoans in this 3-week-old set ranged from $17.1^{\circ}/_{\circ}$ to $36.2^{\circ}/_{\circ}$ with a mean of $24.76^{\circ}/_{\circ}$. The mean total protozoan species in this set was 44.8, and there was a mean for flagellates of 11.3.

Included in Table IV are the results obtained 11 July 1970 from sampling a set of 10 units that had remained in South Fishtail Bay for 9 days. The percentage of flagellates among the colonizing protozoans ranged from 33.3% to 54.3% with a mean of 43.65%. The total number of protozoan species ranged from 28 to 38 with a mean of 32.

North Fishtail Bay is the northern half of the tail of the fish shaped Douglas Lake. It is less densely inhabited, less developed and less used than South Fishtail Bay. North Fishtail Bay is the only habitat in the five-square-mile lake that contains a stand of water lilies. Table V gives the results obtained from sampling three sets of units at different times. The first set which had been exposed in the lake for one year was harvested and sampled 26 June 1974. The mean for ten units was 35.6 protozoan species with an average of 7.4 flagellates or a mean flagellate percentage of 20.8% of the total species. On 4 October 1974, a week after lake overturn, the mean number of protozoan species was 60.9 and the mean of flagellates was 19.9 or 32.7%. Three weeks later a set of units with an exposure of 3 weeks had a mean of 47 total species and 16.1 flagellates for a flagellate percentage of 34.3% of the total number of species. Since flagellates generally have highly successful colonization rates in the early stages of community development, these results suggest that overturn recreates "pioneer conditions" for flagellates in some way. Since the total number of species was also higher after overturn, the increase in flagellate species percentage of the total number of species was not due to reduced competition for space with other major groups of protozoans.

Table VI shows the number of protozoan species, numbers of flagellates, and percentages of flagellates that colonized PF units suspended in an eutrophic pond (Nichol's Bog Pond) weekly over a seven-week period in 1970 and periodically during a seven-week period

Table V. Percentages of flagellates colonizing polyurethane foam units in multiple sets (North Fishtail Bay, Douglas Lake, Michigan)

Exposure	Sampling date	Unit number	Total flagellates	Total protozoan species	% flagellates
•	C Y 1074		12	40	20
1 year	6 June 1974	· . 1	12	40	30
		2	11	42	26.2
		3	9	40	22.5
		4	7	36	19.4
		5	1	24	4.2
		6	8	47	17
		7	7	26	26.9
		8	7	28	25
		9	5	37	13.5
		10	7	36	19.4
1 year	10 April 1974	1	24	77	31.2
(1 week after		2	19	81	23.5
lake overturn)		3	19	57	33.3
		4	15	58	25.9
		5	12	53	22.6
		6	25	60	41.7
		7	27	67	40.3
		8	18	50	36
		9	19	51	37.3
		10	21	55	38.2
3 weeks	25 October 1974	1	18	53	34
(4 weeks after		2	19	50	38
lake overtutn)		3	14	45	31
,		4	18	50	36
		5	12	45	26.7
		6	16	47	34
		7	13	43	30.2
		8	18	45	40
		9	18	49	36.7
		10	15	43	34.9

in 1974. The percentages of flagellates ranged from $33.3^{\circ}/_{0}$ to $62.8^{\circ}/_{0}$ with a mean of $46.5^{\circ}/_{0}$ for all samplings. The mean percentage of flagellates for samples from units that had exposures of two weeks or longer was $46.8^{\circ}/_{0}$ with a range of $41.6^{\circ}/_{0}$ to $54.5^{\circ}/_{0}$. Clearly the percentage of flagellates in the protozoan communities is higher in bog ponds than in Douglas Lake.

Table VII contains the names of the 20 most common species found during the early summer colonization of PF units immersed in Douglas Lake and in Nichol's Bog Pond. The table also shows the orders among which these species were distributed. The species were widely distribut-

Table VI. Percentage of flagellates colonizing polyurethane foam units in eutrophic pond (Nichol's Bog Pond)

Sampling date	Week(s) exposure	Total flagellates	Total protozoan species	Percentage of flagellates
30 June 1970	1	22	35	62.8
7 July 1970	2	18	40	45
14 July 1970	3	28	66	42
21 July 1970	4	. 28	54	51.8
28 July 1970	5	25	56	44.6
4 August 1970	6	25	. 58	43
11 August 1970	. 7	. 30	55	54.5
2 July 1974	1	20	40	50
		16	43	37.2
		15	45	33.3
16 July 1974	3	.20	48	41.6
30 July 1974	5	21	47	44.7
30 August 1974	7	28	52	53.8

Table VII. Twenty most common early summer pioneer and persistant flagellates

Douglas Lake	Nichol's Bog Pond	
Ceratium hirundinella (Stein)	Astasia klebsii Lemmermann	
Chromulina spp. Cienkowsky	Bodo caudatus Dujardin	
Cryptochrysis commutata Pascher	Chlamydomonas spp. Ehrenberg	
Cryptomonas ovata Ehrenberg	Collodictyon triciliatum (Carter)	
Cyathomonas truncata Ehr	Cryptomonas erosa Ehr	
Dinobryon divergens Imhof	Cyathomonas truncata Ehr	
Dinobryon stipitatum Stein	Dinobryon sertularia Ehr	
Entosiphon sulcatum (Dujardin)	Entosiphon sulcatum (Dujardin)	
Mallomonas acaroides Perty	Euglena deses Ehr	
Mallomonas caudata Conrad	Euglena rubra Hardy	
Mallomonas pseudocoronatum Prescott	Gonyostomum semen Dujardin	
Ochromonas ludibunda Pascher	Hemidinium nasutum Stein	
Peranema trichophorum (Ehr)	Lepocinclis ovum (Ehr)	
Peridinium cinctum (Muller)	Massartia musei Schiller	
Peridinium willei Huitfeld-Kass	Notosolenus apocamptus Stokes	
Phacotus lenticularis (Ehr)	Phacus pleuronectes Muller	
Trachelomonas hispida (Stein)	Phacus pyrum (Ehr)	
Trachelomonas volvocina Ehr	Peranema trichophorum (Ehr)	
Rhodomonas lacustris Pascher and Ruttner	Synura uvella (Ehr)	
Uroglenopsis	Trachelomonas hispida (Stein)	

	Distribution by Orders	
	Douglas Lake	Nichol's Bog Pond
Chrysomonads	8	2
Cryptomonads	4	2
Phytomonads	1	1
Euglenoids	4	10
Dinoflagellates	3	2
Protomonads	0	2
Chloromonads	0	1

ed among the orders in both the lake and the pond, but the dominant category was chrysomonds (8 species) in Douglas Lake and euglenoids (10 species) in Nichol's Bog Pond.

4. DISCUSSION

It is rather well documented that protozoans, in general, have broad water quality tolerance ranges (Cairns 1969, Noland, Gojdics 1967, Yongue et al. 1973) even though individual species may be quite sensitive to minor shifts in quality. Depending upon the system and its protozoan communities, major ecological perturbations in the system (e.g., lake overturn; Cairns et al. 1976) may cause significant changes in numbers of colonizing protozoans, but generally the colonization is represented by a characteristic dynamic equilibrium (i.e., oscillations about a mean; Yongue, Cairns 1971). It has been suggested (Yongue, unpublished dissertation) that numbers of species in a system appear to be more closely related to temperature that to pH, dissolved oxygen concentration, and total hardness, The water quality of Douglas Lake, based on pH, dissolved oxygen concentration, and total hardness, was much the same during the early summer period of 1973 and 1974. The water temperature was slightly higher in 1973 (mean <1°C) but this would not seem to account for the notable qualitative difference between the two periods (i.e., the three blooms of photosynthetic organisms in 1974). If these blooms represent significant changes in water quality, the changes apparently are not detectable during pioneer protozoan colonization of PF units if total colonizing species is used for the evaluation.

When the total numbers of colonizing pioneer protozoans were subdivided into the major groups—flagellates, sarcodines, and ciliates—it was apparent in both years (1973 and 1974) that the flagellates very quickly became established components of the communities and that the total numbers of flagellates were relatively constant for the 14 days. When these were plotted as percentages of the total colonizers, a rapidly descending curve that stabilized at approximately $35^{\circ}/_{\circ}$ was generated. Since this was characteristic for both years, the possibility was considered that this pattern might be a general one. To check this possibility, sets of replicated data from other sampling periods were examined.

In a set of units that consisted of 10 replicates that had remained in South Fishtail Bay of Douglas Lake for 2 years and that had been

sampled 28 June 1974, the percentages of flagellates colonizing the units ranged from $13.6^{\circ}/_{\circ}$ to $33.3^{\circ}/_{\circ}$ with a mean of $20.54^{\circ}/_{\circ}$. In a similar set that had remained in that vicinity for 1 year, the percentages of flagellates ranged from $11.1^{\circ}/_{\circ}$ to $30^{\circ}/_{\circ}$ with a mean of $19.07^{\circ}/_{\circ}$. Without question, even for long-term exposures, the percentages of flagellates during early summer (characteristically, a relatively stable period) were considerably below $35^{\circ}/_{\circ}$ and similar to that which might have been predicted from the pioneer colonization study.

Lake overturn significantly increases the number of protozoans colonizing PF units (Cairns et al. 1976, Yongue, Cairns 1974) and to a more limited extent, so do mechanical perturbations (Cairns et al. 1971). Data from a 3-month-old set of 10 units from South Fishtail Bay that was hervested 1 week after fall lake overturn showed this effect. As noted in Tab. IV, during the early summer the number of colonizing protozoans ranged from 45 to 55 species. For this post-overturn set, the mean number of protozoan species was 60.60. The percentages of flagellates in this series ranged from 21.2% to 29.7% with a mean of 25.6%. This percentage was higher than the percentages for the preoverturn sets, suggesting that the overturn causes concomitant redistribution of profundal nutrients and species.

That 14 days are needed in order for a community to become dynamically stable in Douglas Lake at this stage of its life history is supported by the data from a set of units that was harvested after 9 days exposure several years earlier from this same vicinity of South Fishtail Bay. The total numbers of protozoan species were lower than that from the sets previously discussed. The range was from 28 to 38 species with a mean of 32. The percentages of flagellates tend to agree with the results from the pioneer colonization study. These percentages ranged from 33.3% to 54.3% with a mean of 43.65%.

Evidence that there is a qualitative difference associated with the percentages of flagellates among protozoans that colonize PF units is shown in Tab. VII. Douglas Lake, a moderately enriched body of water, tends to have a greater proportion of chrysomonads than flagellates in the other orders while Nichol's Bog Pond, a highly enriched body of water, has a greater proportion of euglenoids.

5. CONCLUSIONS

Pioneer colonization of polyurethane foam units by protozoans is characterized by an increasing trend until equilibrium is reached. Flagellate species are a more dominant portion of the protozoan community in the early stages of colonization than in the latter stages. Equilibrium of flagellate species was reached rather quickly—much before equilibrium was reached by the other major protozoan groups. Lake overturn was followed by an increase in number of flagellate species on the artificial substrates which was true for the other major taxonomic groups as well. This preliminary information suggests either (1) flagellate colonization may be necessary before full colonization of the other groups can occur, or (2) flagellate species have a higher invasion rate than the other major taxonomic groups and thus reach equilibrium conditions much more rapidly.

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